

Automated 4AAP Phenolic Method

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16. ABSTRACT An automated colorimetric method for the determination of phenol in water and wastes is presented. This method is an automated version of the 4AAP method, capable of analyzing from ten to twenty samples per hour. The minimum detectable level is 1 ug. phenol/l.		
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Automated 4AAP Phenolic Method

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Modifying a manufacturer's procedure has produced a method for sampling phenolics that is reportedly comparable in accuracy to the manual method, at a rate approximately nine times greater.

Waste products from oil refineries, coke areas, and chemical plants may contain high concentrations of phenolic materials. These compounds are considered pollutants because they cause unpleasant taste in potable water supplies. Chlorination of water containing phenolics produces chlorophenols, which have even more disagreeable tastes and odors than the parent material. Compliance monitoring of point source discharges, as required by the National Pollution Discharge Elimination System (NPDES), has accelerated the need for a simple method that can detect phenol levels of 1 µg/l, so an evaluation of an automated method¹ for phenolic material to determine its applicability to all types of waters and wastes as well as its comparability to the existing EPA-approved method² was performed.

Since 1951, phenolic concentrations in water and wastewaters have been determined by the manual 4-aminoantipyrine method (4-AAP).³ This method is time consuming, because it requires distillation of a 500-ml sample and subsequent reaction of the phenol in the distillate with ferricyanide and 4AAP. At concentrations above 50 µg/l, the red complex formed can be measured directly; however, at lower levels, the complex must be extracted before the colorimetric measurement. Because of the time required to complete the distillation and the subsequent extraction, most laboratories are able to analyze only 12-18 samples/day.

An automated version of this classic procedure that can be used to analyze 20 samples/hr has been developed.* This task

is accomplished through the use of a much smaller sample for the distillation and elimination of the extraction step. Although a range of 0-50 µg/l was claimed by the company, the author's evaluation of the proposed method indicated a more realistic detection limit of about 50 µg/l. Through changes in the sampling rate, manifold and reagent concentrations, the sensitivity was lowered to 2 µg/l. Following is the modified automated procedure.

Materials and Methods

Apparatus. Technicon AutoAnalyzer (I or II), or equivalent, consisting of sampler, manifold, proportioning pump II or III, heating bath with distillation coil, distillation head, colorimeter equipped with 50-mm flow cell and 505 or 520-nm filter, and recorder.

Reagents. The distillation reagent is produced by adding 100 ml of conc. phosphoric acid (85 per cent H₃PO₄) to 800 ml of distilled water, cooling, and diluting to 1 l. Sodium hydroxide (1 N). Forty g sodium hydroxide is dissolved in 500 ml of distilled water, cooled, and diluted to 1 liter.

To produce the buffered potassium ferricyanide, 2.0 g potassium ferricyanide, 3.10 g boric acid and 3.75 g potassium chloride are dissolved in 800 ml of distilled water. The pH is adjusted to 10.3 with 1 N sodium hydroxide and the solution is diluted to 1 l. Then 0.5 ml of Brij-35† is added.

The solution of 4-Aminoantipyrine is produced by dissolving 0.65 g of 4-aminoantipyrine in 800 ml of distilled water and diluting to 1 l.

By dissolving 1.1 g ferrous ammonium sulfate in 500 ml distilled water containing 1 ml H₂SO₄ and diluting to 1 l with freshly boiled and cooled distilled water, the ferrous ammonium sulfate solution is produced.

For the stock phenol, 1.00 g phenol is dissolved in 500 ml of distilled water and diluted to 1.0 liter. One g copper sulfate and 0.5 ml conc. phosphoric acid are added as preservatives. One ml of this solution contains 1.0 mg phenol. By diluting 10.0 ml of stock phenol solution to 1 l, standard phenol solution A is produced. One ml of solution A contains 0.01 mg phenol.

Standard phenol solution B is produced by diluting 100.0 ml of standard phenol solution A to 1 l with distilled water; 1.0 ml of this solution contains 0.001 mg phenol.

A further dilution of 100.0 ml of standard phenol solution B to 1 l with distilled water produces standard solution C, of which 1.0 ml contains 0.0001 mg phenol.

The following working standards are prepared by diluting the indicated amounts of the appropriate standard solution to 100 ml with distilled water. Each working standard should be preserved by adding 0.1 g copper sulfate and 2 drops of conc. phosphoric acid to 100 ml.

Standard Solution—ml	Phenol Concentration—µg/l
Solution C:	
1.0	1.0
2.0	2.0
3.0	3.0
5.0	5.0
Solution B:	
1.0	10.0
2.0	20.0
5.0	50.0
10.0	100.0
Solution A:	
2	200
3	300
5	500

Procedure. The manifold is set up as shown in Fig. 1 or 2. One should fill the wash receptacle by siphon, using Kel-F tubing with a fast flow (1 l/hr).

The colorimeter and recorder should warm up for 30 min. One should run a baseline with all reagents, feeding distilled water through the sample line. Polyethylene tubing should be used for the sample line and glass test tubes for standards and samples. When new tubing is used, approximately 2 hrs may be required to remove residual phenol from the tubing and obtain a stable baseline. Appropriate phenol standards should be placed in the sampler in order of decreasing concentration and the loading of the sampler tray completed by filling it with unknown samples. After a stable baseline has been obtained, the sampler is started. It should be noted that if samples have not been preserved, it is necessary to add 0.1 g copper sulfate and 2 drops of conc. phosphoric acid to 100 ml of the sample.

Calculation. A standard curve is prepared by plotting peak heights of standards against concentration values. The concentration of the samples is then computed by comparing sample peak heights with the standard curve.

Discussion

In the Technicon method, the wash receptacle is filled by pumping distilled water through Tygon tubing, but when this system was used, a high background was obtained, and the low standards were not readable. This background contamination, which is probably a phenolic material, was eliminated by filling the wash receptacle by siphon (using glass or Kel-F tubing). A similar interference, noted when a pump tube was replaced, was eliminated by pumping reagent or distilled water through the tube for about 2 hr. Finally, the resample line flow was increased from 1.0 to 1.2 ml/min., and the waste line flow from the still was reduced from 1.2 to 1.0 ml/min. These changes resulted in a smooth, stable baseline. Flow diagrams are shown in Fig. 1 and 2. The distillation system of this method was found to be very satisfactory.

The Technicon method recommends a

*Registered trademark of Atlas Chemical Industries, Inc., available from Technicon Instruments Corp.

*By Technicon Instruments Corp.

sampling rate of 40 samples/hr (4:1). At this sampling rate, it is very difficult to identify sample peaks, especially when a sample of high phenol concentration is followed by one at a low concentration (see Fig. 3). This system was improved by reducing the sampling rate to 20/hr. At this slower sampling rate, a longer wash is provided, and sample peaks are readily identified (see Fig. 4). Though a reduction in the sampling rate gave a better resolution of the sample peaks, it did not improve the sensitivity.

Reagents. The reagent 4-aminoantipyrine reacts with phenol in the presence of potassium ferricyanide at a pH of 10 to form a stable reddish-brown colored antipyrine dye. The manual phenol method uses a ratio of 4:1 (potassium ferricyanide to 4-aminoantipyrine) for color development. The Technicon system uses a buffered solution that contains 1.0 g/l of potassium ferricyanide and a solution of 1.0 g/l 4AAP, both of which are added to the distillate for color development. This ratio of 1:1 ferricyanide to 4AAP produces the typical brownish-red color with phenolic materials, but the method is not sensitive below 50 $\mu\text{g/l}$. Greater sensitivity was achieved when the concentration of potassium ferricyanide solution was increased from 1 to 3 g/l. Peaks obtained with the 1:1 ratio and the 3:1 ratio are shown in Fig. 5. Specifically, the peak height of the 100 $\mu\text{g/l}$ standard increased from 29 to 53 units, the 10 $\mu\text{g/l}$ standard increased from 3.5 to 6 units, and the 2 $\mu\text{g/l}$ increased from 1.5 to 2 units.

In the Technicon method, the stock standard phenol solution is prepared by dissolving phenol in 0.05 N hydrochloric acid. In the original manual procedure for phenolics, hydrochloric acid was not used for distillation because it would distill over, resulting in a very acid distillate. Phosphoric acid was chosen because it is nonvolatile and noncharring. Therefore, the stock phenol solution was prepared by dissolving 1.0 g of phenol in 1 l distilled water containing 1 g of copper sulfate and 0.5 ml of conc. phosphoric acid. This solution is stable. The working standards are also stable for 1 week when 0.1 g of copper sulfate and 2 drops of phosphoric acid are added to 100 ml. All samples must contain copper sulfate and phosphoric acid.

The interferences of this automated method are the same as with the manual method. Interferences from chlorine, sulfur compounds, oils, and tars are eliminated by pretreatment of the sample as suggested in *Standard Methods*.³

Results

The precision of this method was determined at four separate concentration levels for two working ranges (2-100 $\mu\text{g/l}$ and 5-500 $\mu\text{g/l}$). The concentrations included a level near the lower sensitivity level of the method, two intermediate concentrations, and one near the upper limit. For phenol concentrations of 4, 15, 43 and 89 $\mu\text{g/l}$, the

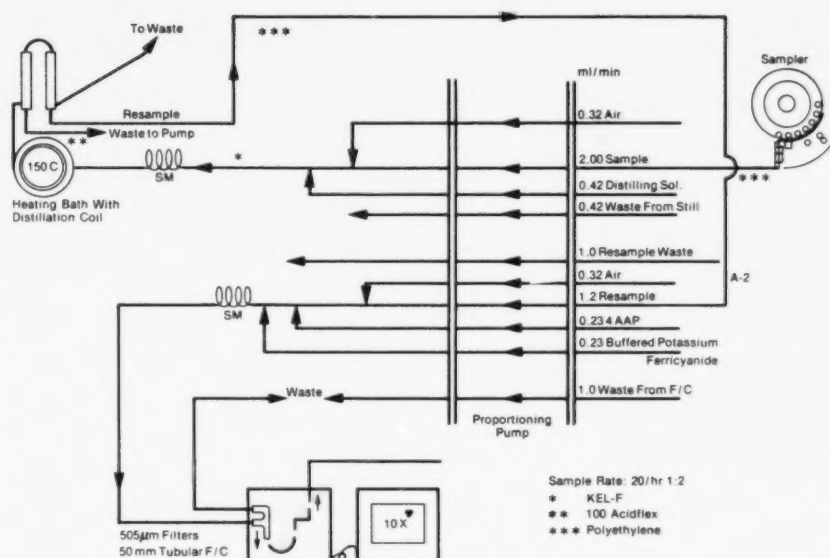


Fig. 1 Flow Set-up for Analyzer I

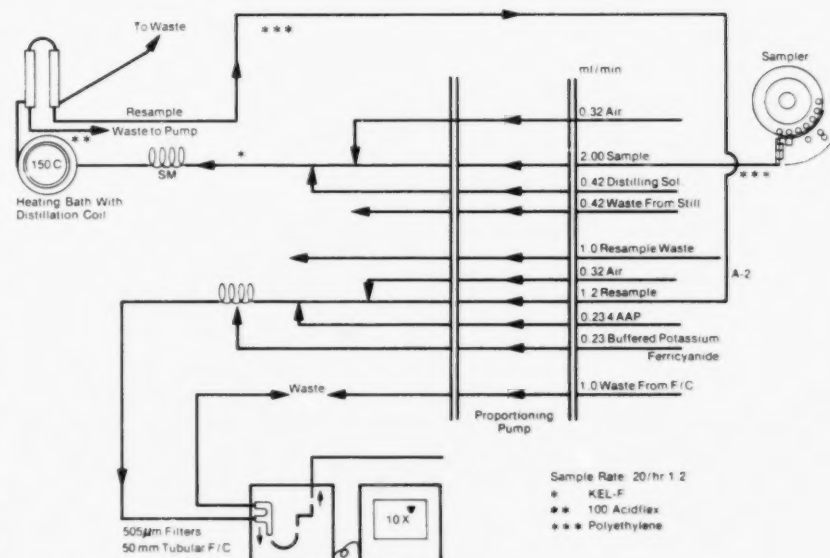


Fig. 2 Flow Set-up for Analyzer II

standard deviations were ± 0.5 , ± 0.6 , ± 0.6 , and ± 1.0 $\mu\text{g/l}$, respectively. At concentrations of 73, 146, 299, and 447, the standard deviations were ± 1.0 , ± 1.8 , ± 4.2 , and ± 5.3 $\mu\text{g/l}$, respectively. The samples used for these determinations were prepared by adding known amounts of phenol standard to a treated domestic/industrial waste effluent sample.

The per cent recovery was determined at two different levels for each working range

by spiking the waste sample with four concentrations of phenol. At concentrations of 5, 83, 168, and 490 $\mu\text{g/l}$, the recoveries were 72, 98, 97 and 98 per cent respectively (Table I). Recovery was also determined by comparing results obtained for sewage and petroleum waste by the automated method to those obtained by the manual method. Results obtained by the automated method were 3-4 per cent lower than those obtained by the manual method (Table 2).

TABLE 1
Recovery of Phenol from Sewage Samples

Sample	Phenol in Sample $\mu\text{g/l}$	Phenol Added $\mu\text{g/l}$	Phenol Found $\mu\text{g/l}$	Recovery per cent
1	4	3	5	72
2	43	40	82	99
3	73	100	168	97
4	300	200	490	98

TABLE 2
Comparison of Results Obtained by the Automated and Manual Method

Method	Sample	Phenol in Sample $\mu\text{g/l}$
Manual	sewage	76
Automated	sewage	73
Manual	petroleum waste	6.2
Automated	petroleum waste	6.0

TABLE 3
Recovery of Phenolics

Phenolics	Conc. found $\mu\text{g/l}^*$
o-cresol	65
m-cresol	69
p-cresol	2
2-naphthol	3
3-4-dimethylphenol	2
p-chlorophenol	59
o-chlorophenol	59
4-trichlorophenol	30
m-aminophenol	< 2
o-aminophenol	3
p-aminophenol	6
p-nitrophenol	4
o-nitrophenol	4
2-amino-4-nitrophenol	< 2

*Original concentration 100 $\mu\text{g/l}$

The recovery of substituted phenolics at 100 $\mu\text{g/l}$ was also determined. Recoveries ranged from 2-69 per cent (Table 3). These results indicate that values obtained by this method, as with the manual method, are highly dependent on the type of phenolic material present in the sample.¹

Conclusions

The results obtained with the automated phenol method are comparable to those obtained by the manual phenol method. The sensitivity as well as the precision and accuracy of the method are satisfactory for the determination of phenol in surface waters and in domestic and industrial wastes. The precision of this method is not dependent on the skill of the analyst, as it is with the manual method; recovery is comparable to that of the manual method. Since at least 20 samples/hr can be analyzed by the automated system, it offers a distinct advantage over the manual procedure. As in the manual method, recovery of substituted phenolics is dependent on the type and extent of substitution.

It is not the authors' intent to suggest that

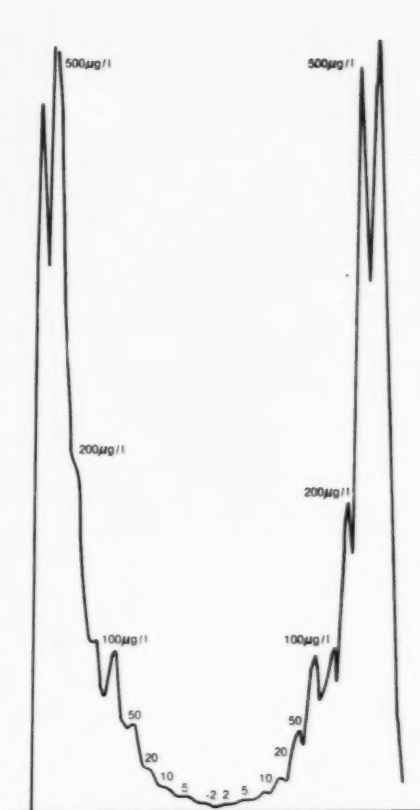


Fig. 3 Sample Peaks at a Sampling Rate of 40/hr (4:1) 1 g/l Potassium Ferricyanide, 0-500 $\mu\text{g/l}$ phenol

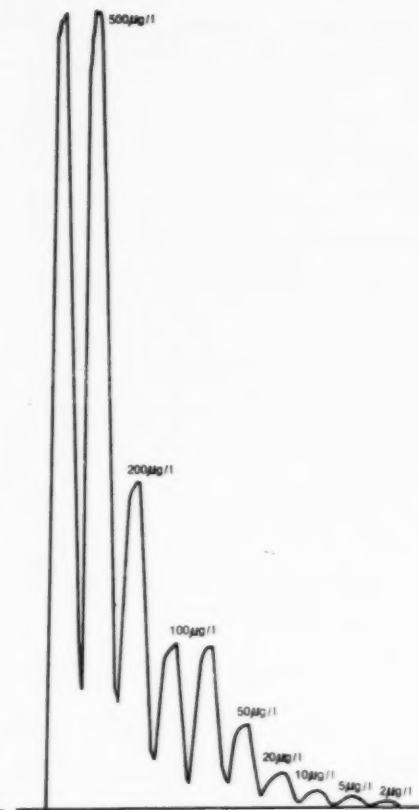


Fig. 4 Sample Peaks at a Sampling Rate of 20/hr (2:1) 1 g/l Potassium Ferricyanide, 0-500 $\mu\text{g/l}$ phenol

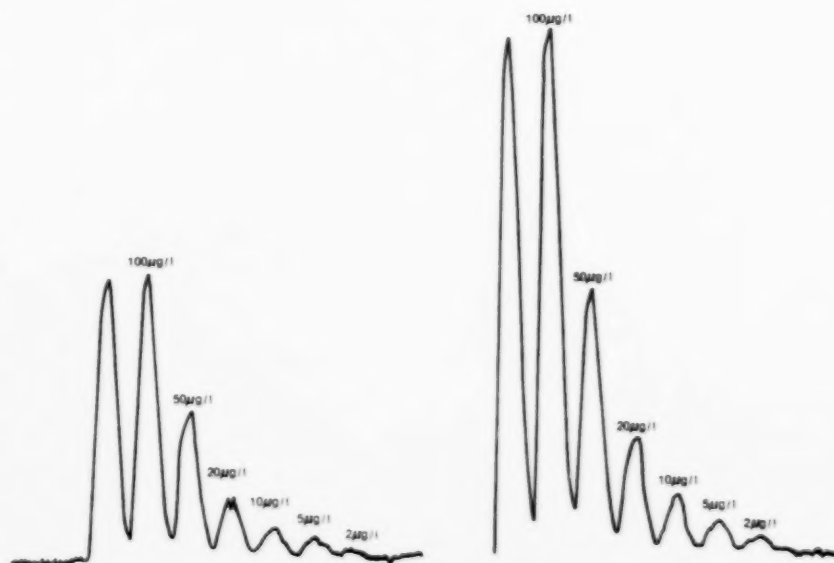


Fig. 5 Comparison of Sample Peaks at a Sampling Rate of 20/hr—1 g/l and 3 g/l Potassium Ferricyanide

this automated version replace the manual method. It is offered as an alternate procedure to those laboratories faced with large numbers of samples for analysis. The interferences of this automated version are the same as those encountered with the manual method, but it does have the advantage of displaying possible interferences through the use of a recorder. The manual method can serve as a referee method and will still be used by those laboratories not requiring the ability to analyze 20 samples per hour.

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